EVIDENCE ON THE ADAPTATION OF PARAMÆCIA TO DIFFERENT ENVIRONMENTS.

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The fact being established that my pedigree culture of *Paramœcium aurelia* (I.) undoubtedly has unlimited power of reproduction without conjugation or artificial stimulation,[1] a culture of *Paramœcium caudatum* was started for comparison, in order to determine if this animal would show throughout its life history characteristics of specific value and also to determine if it would continue to live and reproduce indefinitely without conjugation or artificial stimulation.

The results with this culture led me to conclude, as did Jennings and Hargitt,[2] that *caudatum* is a distinct species. This point I have discussed in a previous paper.[3] The results in regard to the second point are briefly presented at this time.

The pedigree culture of *Paramœcium caudatum* (X.) was started on May 14, 1910, and has been continued under observation to the present time, December 1, 1911. The methods employed have already been described in detail in earlier papers on pedigree cultures of Infusoria. It is only necessary to state here that the culture was begun by placing a large "wild" individual on a depression slide in about five drops of culture medium. When this individual had divided twice, producing four animals, each of these was placed on a separate slide, forming the four lines of the culture. Thereafter (until June 1, 1911) a single cell from each of the lines was isolated daily in fresh culture medium and the number of divisions during the previous twenty-four hours was recorded.

In regard to the culture of *Paramœcium aurelia* (I.), which

[1] L. L. Woodruff: "Two Thousand Generations of *Paramœcium*." *Archiv für Protistenkunde*, Bd. 21, 3, 1911.

[2] H. S. Jennings and G. T. Hargitt: "Characteristics of the Diverse Races of *Paramœcium*." *Journ. Morph.*, Vol. 21, no. 4, 1910.

[3] L. L. Woodruff: "*Paramœcium aurelia* and *Paramœcium caudatum*." *Journ. Morph.*, Vol. 22, no. 2, 1911.

served as a control and for comparison with the *P. caudatum* culture, there are no results to be recorded which are not in entire agreement with these already published. The culture has kept on the even tenure of its way and is now, after over four and one half years of daily observation, at the 2,705th generation, and in every way in as normal morphological and physiological condition as at the start. Given a favorable environment, this race clearly has unlimited power of reproduction without conjugation or artificial stimulation.

The pedigree culture of *Paramœcium caudatum* (X.), which was subjected from the start to the 500th generations (twelve and one half months) to identically the same treatment and culture medium as the *P. aurelia* culture, showed during the first 350 generations (eight months) essentially the same rate of reproduction as the *aurelia* culture. However, an examination of the data (cf. Figs. 1 and 2) shows that a slow decline in division rate set in at the start which finally resulted in a race of cells possessing many of the morphological and physiological characteristics described by Calkins[1] in his careful study of pure lines of this species of *Paramœcium*. After about the 450th generation it became increasingly difficult to keep the animals alive on the slides in the culture medium which was supplied fresh daily. However, the cells left over from the daily isolations, which were allowed to accumulate in the old culture liquid, appeared healthy and continued to reproduce slowly. If these were transferred again to fresh medium they would divide a few times and then die. Finally they would not live twenty-four hours in the fresh medium.

By substituting from the "stock" in this way, the direct lines of the culture were kept replenished for nearly three months; but finally it was evident that it was impossible to continue the culture by this method, so that the exact number of generations could be determined, and accordingly, at the 500th generation, the method was abandoned, and the animals were thereafter carried in small flasks of old infusions, *i. e.*, they were bred in a comparatively large volume of the same type of medium to which

[1] G. N. Calkins: "The Life Cycle of *Paramœcium caudatum*." *Archiv für Entwickelungsmechanik der Organismen*, Bd. 15, 1, 1902. "Death of the A Series of *Paramœcium caudatum*. Conclusions." *Journ. Exper. Zoöl.*, Vol. 1, no. 3, 1904.

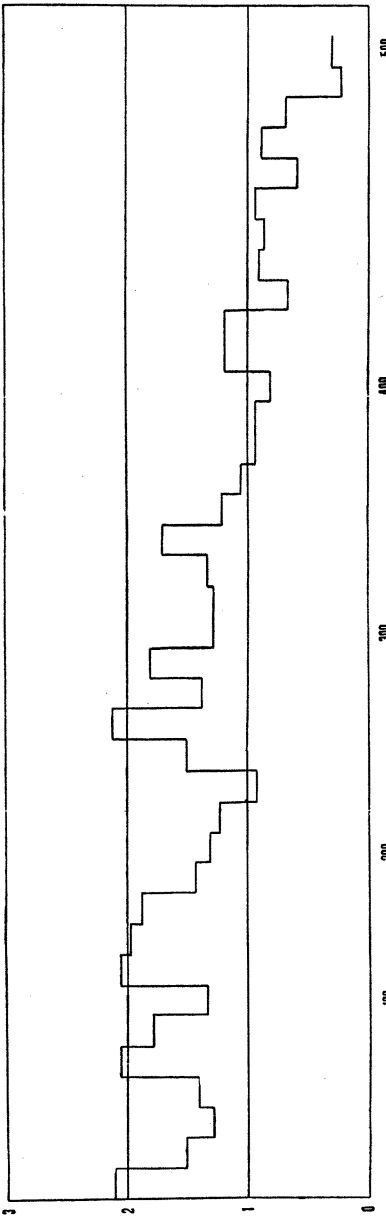

FIG. 1. *Paramæcium caudatum* (Culture X.). Graph of the rate of reproduction for the first 500 generations (May 14, 1910 to June 1, 1911). See text.

The average rate of division of the four lines of the culture is again averaged for *ten-day* periods. The figures 100, 200, etc., represent generations and are placed below the ten-day periods in which they were attained.

they had been previously subjected—but a medium which was from several days to several weeks old.

Under these conditions this culture of *P. caudatum* now flourishes, and it is continued by isolating a few cells every few weeks and inoculating with them another small flask of old infusion. Under these conditions it is impossible, of course, to determine with accuracy the rate of division or the number of generations attained to date, but the organisms are apparently in a normal physiological condition. However, it is still im-

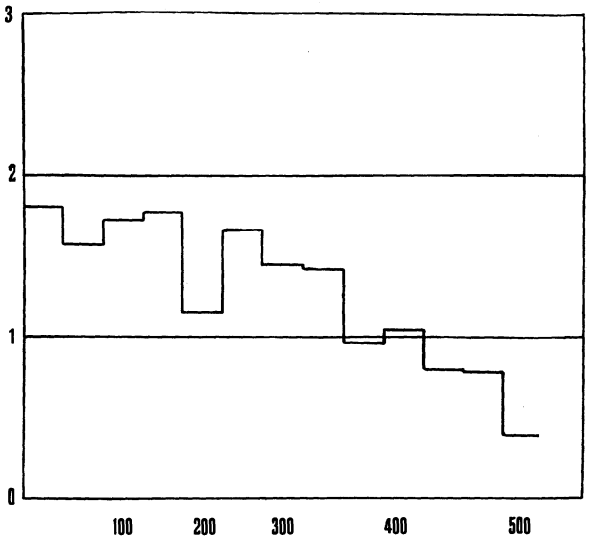

FIG. 2. *Paramœcium caudatum* (Culture X.). Graph of the rate of reproduction for the first 500 generations (May 14, 1910, to June 1, 1911). See text.

The average rate of division of the four lines of the culture is again averaged for periods of *one month*. The figures 100, 200, etc., represent generations and are placed below the months in which they were attained.

possible to keep them alive on slides in the regulation five drops of fresh culture medium, which has proved so highly favorable for the *aurelia* culture.

Now the question arises: Have the cells conjugated in the larger volume of medium and so been "rejuvenated." Since I have been unable to isolate the animals each day, I cannot *prove* that conjugation has not occurred, for it is possible that one or a few pairs have conjugated unobserved and have given rise to the

present generations. The only way to prove that conjugation has not occurred is to make the conditions such that it is an impossibility for it to occur, *i. e.*, by *daily isolations and record of generations*. Since the physiological condition of this pedigree culture prohibited this after the 500th generation, I have adopted the method employed by many investigators of problems of this nature and allowed the Infusoria to accumulate in considerable numbers. I have, however, in order to increase the accuracy of the method, confined the cells in as small a volume of old infusion as possible and have examined the flasks at frequent intervals for signs of conjugation. I have never seen a single pair of conjugants in all the multitude of cells which I have examined, and it seems *highly improbable* that conjugation has occurred. It should be emphasized that, if conjugation has taken place, it has not so altered the physiological condition of the cells that they will live under the slide method of culture.

This culture, then, is apparently in as healthy a condition as at the beginning of the work, but it has become so modified that the animals are unable to exist in small volumes of fresh infusions. This is a decidedly interesting result in the light of the work of other investigators on *Paramœcium caudatum*, since it shows that a race of cells may exhibit all the signs of "senile degeneration" at the end of a typical "cycle" of generations, and still may appear healthy and exhibit a normal rate of reproduction when put under other conditions which approximate what is probably the usual environment of wild paramœcia.

In other words, this culture of *P. caudatum* substantiates the conclusion of Calkins that, under the conditions of his experiments, this organism may pass through a "cycle" which finally terminates in death; but it further shows that this "cycle" is probably an artificial one which is brought about by the subjection of the race to an environment which is not suitable for its prolonged existence. This culture also shows that pure lines of different species of *Paramœcium* (*aurelia* and *caudatum*) are adapted to different environmental conditions, in view of the fact that the race of *P. aurelia* has thrived indefinitely on the same culture medium which has proved increasingly unfavorable for the race of *P. caudatum*. It may be that this is actually a specific

difference, but I believe that the fact that these two races belong to different species is merely an incident and that it will be found to be equally a variation of different pure races of the same species as the results of Jennings clearly indicate.[1]

Conclusions.

1. The discrepant results of various workers on the longevity of paramæcia is in all probability due to variations in the cultural demands of the races isolated for study.

2. It is probable that most, if not all, normal individuals have, under suitable environmental conditions, unlimited power of reproduction without conjugation or artificial stimulation.

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[1] Jennings and Hargitt: *loc. cit.*, p. 538. Jennings: *Amer. Naturalist*, Vol. 45, p. 83, 1911.